

# Substitute Sequence Listing

<110> VERMEIJ, Paul

<120> Hybrid toxins comprising shiga or shiga-like toxin subunits fused to escherichia coli heat labile enterotoxin subunits and vaccines thereof

<130> I-2003.006 us

<150> PCT/EP2004/051522

<151> 2004-07-16

<150> EP 03077266.9

<151> 2003-07-21

<160> 4

<170> PatentIn version 3.3

<210> 1

<211> 1325

<212> DNA

<213> Escherichia coli

<220>

<221> CDS

<222> (1)..(954)

<400> 1

atg atg aag tgt ata ttg tta aag tgg ata ctg tgt ctg tta ctg ggt	48
Met Met Lys Cys Ile Leu Leu Lys Trp Ile Leu Cys Leu Leu Leu Gly	
1 5 10 15	
ttt tct tcg gta tcc tat tcc cag gag ttt acg ata gac ttt tcg act	96
Phe Ser Ser Val Ser Tyr Ser Gln Glu Phe Thr Ile Asp Phe Ser Thr	
20 25 30	
caa caa agt tat gta tct tcg tta aat agt ata cgg aca gtg ata tcg	144
Gln Gln Ser Tyr Val Ser Ser Leu Asn Ser Ile Arg Thr Val Ile Ser	
35 40 45	
acc cct ctt gaa cat ata tct cag gga gct aca tcg gta tcc gtt att	192
Thr Pro Leu Glu His Ile Ser Gln Gly Ala Thr Ser Val Ser Val Ile	
50 55 60	
aat cat aca cca cca gga agt tat att tcc gta ggt ata cga ggg ctt	240
Asn His Thr Pro Pro Gly Ser Tyr Ile Ser Val Gly Ile Arg Gly Leu	
65 70 75 80	
gat gtt tat cag gag cgt ttt gac cat ctt cgt ctg att att gaa cga	288
Asp Val Tyr Gln Glu Arg Phe Asp His Leu Arg Leu Ile Ile Glu Arg	
85 90 95	
aat aat tta tat gtg gct gga ttt gtt aat acg aca aca aat act ttc	336
Asn Asn Leu Tyr Val Ala Gly Phe Val Asn Thr Thr Thr Asn Thr Phe	
100 105 110	
tac aga ttt tca gat ttt gca cat ata tca ttg ccc ggt gtg aca act	384
Tyr Arg Phe Ser Asp Phe Ala His Ile Ser Leu Pro Gly Val Thr Thr	
115 120 125	
att tcc atg aca acg gac agc agt tat acc act ctg caa cgt gtc gca	432

Substitute Sequence Listing															
Ile	Ser	Met	Thr	Thr	Asp	Ser	Ser	Tyr	Thr	Thr	Leu	Gln	Arg	Val	Ala
130						135					140				
gcg	ctg	gaa	cgt	tcc	gga	atg	caa	atc	agt	cgt	cac	tca	ctg	gtt	tca
Ala	Leu	Glu	Arg	Ser	Gly	Met	Gln	Ile	Ser	Arg	His	Ser	Leu	Val	Ser
145					150					155					160
tca	tat	ctg	gcg	tta	atg	gag	ttc	agt	ggt	aat	aca	atg	acc	aga	gat
Ser	Tyr	Leu	Ala	Leu	Met	Glu	Phe	Ser	Gly	Asn	Thr	Met	Thr	Arg	Asp
				165					170					175	
gca	tca	aga	gca	gtt	ctg	cgt	ttt	gtc	act	gtc	aca	gca	gaa	gcc	tta
Ala	Ser	Arg	Ala	Val	Leu	Arg	Phe	Val	Thr	Val	Thr	Ala	Glu	Ala	Leu
			180					185					190		
cgg	ttc	agg	caa	ata	cag	aga	gaa	ttt	cgt	ctg	gca	ctg	tct	gaa	act
Arg	Phe	Arg	Gln	Ile	Gln	Arg	Glu	Phe	Arg	Leu	Ala	Leu	Ser	Glu	Thr
		195					200					205			
gct	cct	gtt	tat	acg	atg	acg	ccg	gaa	gac	gtg	gac	ctc	act	ctg	aac
Ala	Pro	Val	Tyr	Thr	Met	Thr	Pro	Glu	Asp	Val	Asp	Leu	Thr	Leu	Asn
	210					215					220				
tgg	ggg	aga	atc	agc	aat	gtg	ctt	ccg	gag	tat	cgg	gga	gag	gct	ggt
Trp	Gly	Arg	Ile	Ser	Asn	Val	Leu	Pro	Glu	Tyr	Arg	Gly	Glu	Ala	Gly
225					230					235					240
gtc	aga	gtg	ggg	aga	ata	tcc	ttt	aat	aat	ata	tca	gcg	ata	ctt	ggt
Val	Arg	Val	Gly	Arg	Ile	Ser	Phe	Asn	Asn	Ile	Ser	Ala	Ile	Leu	Gly
				245					250					255	
act	gtg	gcc	gtt	ata	ctg	aat	tgt	gga	aat	tca	tca	aga	aca	atc	aca
Thr	Val	Ala	Val	Ile	Leu	Asn	Cys	Gly	Asn	Ser	Ser	Arg	Thr	Ile	Thr
			260					265					270		
ggt	gat	act	tgt	aat	gag	gag	acc	cag	aat	ctg	agc	aca	ata	tat	ctc
Gly	Asp	Thr	Cys	Asn	Glu	Glu	Thr	Gln	Asn	Leu	Ser	Thr	Ile	Tyr	Leu
		275					280					285			
agg	gaa	tat	caa	tca	aaa	gtt	aag	agg	cag	ata	ttt	tca	gac	tat	cag
Arg	Glu	Tyr	Gln	Ser	Lys	Val	Lys	Arg	Gln	Ile	Phe	Ser	Asp	Tyr	Gln
	290					295					300				
tca	gag	gtt	gac	ata	tat	aac	aga	att	cgg	gat	gaa	tta	tga		
Ser	Glu	Val	Asp	Ile	Tyr	Asn	Arg	Ile	Arg	Asp	Glu	Leu			
305					310					315					
ataaagtaaa atgttatgtt ttattttacgg cgttactatc ctctctatat gcacacggag															
ctccccagac tattacagaa ctatgttcgg aatatcgcaa cacacaaata tatacgataa															
atgacaagat actatcatat acggaatcga tggcaggcaa aagagaaatg gttatcatta															
catttaagag cggcgaaaca tttcaggtcg aagtcccgagg cagtcaacat atagactccc															
agaaaaaagc cattgaaagg atgaaggaca cattaagaat cacatatctg accgagacca															
aaattgataa attatgtgta tggaataata aaacccccaa ttcaattgcg gcaatcagta															
tgaaaaacta g															

# Substitute Sequence Listing

<211> 317

<212> PRT

<213> Escherichia coli

<400> 2

Met Met Lys Cys Ile Leu Leu Lys Trp Ile Leu Cys Leu Leu Leu Gly  
1 5 10 15

Phe Ser Ser Val Ser Tyr Ser Gln Glu Phe Thr Ile Asp Phe Ser Thr  
20 25 30

Gln Gln Ser Tyr Val Ser Ser Leu Asn Ser Ile Arg Thr Val Ile Ser  
35 40 45

Thr Pro Leu Glu His Ile Ser Gln Gly Ala Thr Ser Val Ser Val Ile  
50 55 60

Asn His Thr Pro Pro Gly Ser Tyr Ile Ser Val Gly Ile Arg Gly Leu  
65 70 75 80

Asp Val Tyr Gln Glu Arg Phe Asp His Leu Arg Leu Ile Ile Glu Arg  
85 90 95

Asn Asn Leu Tyr Val Ala Gly Phe Val Asn Thr Thr Thr Asn Thr Phe  
100 105 110

Tyr Arg Phe Ser Asp Phe Ala His Ile Ser Leu Pro Gly Val Thr Thr  
115 120 125

Ile Ser Met Thr Thr Asp Ser Ser Tyr Thr Thr Leu Gln Arg Val Ala  
130 135 140

Ala Leu Glu Arg Ser Gly Met Gln Ile Ser Arg His Ser Leu Val Ser  
145 150 155 160

Ser Tyr Leu Ala Leu Met Glu Phe Ser Gly Asn Thr Met Thr Arg Asp  
165 170 175

Ala Ser Arg Ala Val Leu Arg Phe Val Thr Val Thr Ala Glu Ala Leu  
180 185 190

Arg Phe Arg Gln Ile Gln Arg Glu Phe Arg Leu Ala Leu Ser Glu Thr  
195 200 205

Ala Pro Val Tyr Thr Met Thr Pro Glu Asp Val Asp Leu Thr Leu Asn  
210 215 220

Trp Gly Arg Ile Ser Asn Val Leu Pro Glu Tyr Arg Gly Glu Ala Gly  
Page 3

# Substitute Sequence Listing

225 230 235 240

Val Arg Val Gly Arg Ile Ser Phe Asn Asn Ile Ser Ala Ile Leu Gly  
245 250 255

Thr Val Ala Val Ile Leu Asn Cys Gly Asn Ser Ser Arg Thr Ile Thr  
260 265 270

Gly Asp Thr Cys Asn Glu Glu Thr Gln Asn Leu Ser Thr Ile Tyr Leu  
275 280 285

Arg Glu Tyr Gln Ser Lys Val Lys Arg Gln Ile Phe Ser Asp Tyr Gln  
290 295 300

Ser Glu Val Asp Ile Tyr Asn Arg Ile Arg Asp Glu Leu  
305 310 315

<210> 3  
<211> 1325  
<212> DNA  
<213> Escherichia coli

<220>  
<221> CDS  
<222> (951)..(1322)

<400> 3  
atgatgaagt gtatattggt aaagtggata ctgtgtctgt tactggggtt ttcttcggtta 60  
tcctattccc aggagtttac gatagacttt tcgactcaac aaagttatgt atcttcggtta 120  
aatagtatac ggacagtgat atcgaccctt cttgaacata tatctcaggg agctacatcg 180  
gtatccggtta ttaatcatac accaccagga agtttatatt ccgtaggtat acgagggtt 240  
gatgtttatc aggagcggtt tgaccatctt cgtctgatta ttgaacgaaa taatttatat 300  
gtggctggat ttgttaatac gacaacaaat actttctaca gattttcaga ttttgcacat 360  
atatcattgc ccggtgtgac aactatttcc atgacaacgg acagcagtta taccactctg 420  
caacgtgtcg cagcgctgga acgttccgga atgcaaatca gtcgtcactc actggtttca 480  
tcatatctgg cggttaatgga gttcagtggt aatacaatga ccagagatgc atcaagagca 540  
gttctgcgtt ttgtcactgt cacagcagaa gccttacggt tcaggcaaata acagagagaa 600  
tttcgtctgg cactgtctga aactgctcct gtttatacga tgacgccgga agacgtggac 660  
ctcactctga actgggggag aatcagcaat gtgcttccgg agtatcgggg agaggctggt 720  
gtcagagtgg ggagaatatc ctttaataat atatcagcga tacttggtac tgtggccggt 780  
atactgaatt gtggaaattc atcaagaaca atcacaggtg atacttgtaa tgaggagacc 840  
cagaatctga gcacaatata tctcagggaa tatcaatcaa aagttaagag gcagatattt 900

# Substitute Sequence Listing

tcagactatc agtcagaggt tgacatatat aacagaattc gggatgaatt atg aat 956  
Met Asn  
1

aaa gta aaa tgt tat gtt tta ttt acg gcg tta cta tcc tct cta tat 1004  
Lys Val Lys Cys Tyr Val Leu Phe Thr Ala Leu Leu Ser Ser Leu Tyr  
5 10 15

gca cac gga gct ccc cag act att aca gaa cta tgt tcg gaa tat cgc 1052  
Ala His Gly Ala Pro Gln Thr Ile Thr Glu Leu Cys Ser Glu Tyr Arg  
20 25 30

aac aca caa ata tat acg ata aat gac aag ata cta tca tat acg gaa 1100  
Asn Thr Gln Ile Tyr Thr Ile Asn Asp Lys Ile Leu Ser Tyr Thr Glu  
35 40 45 50

tcg atg gca ggc aaa aga gaa atg gtt atc att aca ttt aag agc ggc 1148  
Ser Met Ala Gly Lys Arg Glu Met Val Ile Ile Thr Phe Lys Ser Gly  
55 60 65

gaa aca ttt cag gtc gaa gtc ccg ggc agt caa cat ata gac tcc cag 1196  
Glu Thr Phe Gln Val Glu Val Pro Gly Ser Gln His Ile Asp Ser Gln  
70 75 80

aaa aaa gcc att gaa agg atg aag gac aca tta aga atc aca tat ctg 1244  
Lys Lys Ala Ile Glu Arg Met Lys Asp Thr Leu Arg Ile Thr Tyr Leu  
85 90 95

acc gag acc aaa att gat aaa tta tgt gta tgg aat aat aaa acc ccc 1292  
Thr Glu Thr Lys Ile Asp Lys Leu Cys Val Trp Asn Asn Lys Thr Pro  
100 105 110

aat tca att gcg gca atc agt atg aaa aac tag 1325  
Asn Ser Ile Ala Ala Ile Ser Met Lys Asn  
115 120

<210> 4  
<211> 124  
<212> PRT  
<213> Escherichia coli

<400> 4

Met Asn Lys Val Lys Cys Tyr Val Leu Phe Thr Ala Leu Leu Ser Ser  
1 5 10 15

Leu Tyr Ala His Gly Ala Pro Gln Thr Ile Thr Glu Leu Cys Ser Glu  
20 25 30

Tyr Arg Asn Thr Gln Ile Tyr Thr Ile Asn Asp Lys Ile Leu Ser Tyr  
35 40 45

Thr Glu Ser Met Ala Gly Lys Arg Glu Met Val Ile Ile Thr Phe Lys  
50 55 60

ser Gly Glu Thr Phe Gln val Glu val Pro Gly Ser Gln His Ile Asp  
65 70 75 80

### Substitute Sequence Listing

Ser Gln Lys Lys Ala Ile Glu Arg Met Lys Asp Thr Leu Arg Ile Thr  
85 90 95

Tyr Leu Thr Glu Thr Lys Ile Asp Lys Leu Cys Val Trp Asn Asn Lys  
100 105 110

Thr Pro Asn Ser Ile Ala Ala Ile Ser Met Lys Asn  
115 120